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Forest Insect Laboratory
Coeur d'Alene, Idaho

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TITLE

SUMMARY REPORT OF LOW TEMPERATURE STUDIES 1936 - 1942

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Forest Insect Laboratory
Coeur d'Alene, Idaho

February 24, 1943

To: F. C. Craighead, in Charge, Forest Insect Investigations

From: James C. Evenden, in Charge, Coeur d'Alene Laboratory

Subject: Summary Report of Low Temperature Studies, 1936 - 1942

I am enclosing a copy of my summary report of low temperature studies. This report included those phases of the work conducted at this laboratory which bear directly upon the practical demands of this problem. As stated in the introduction, although I can not consider that all questions concerning the effects of low temperatures upon overwintering larvae of the mountain pine beetle have been answered, I do believe that the practical phases of this problem have been satisfied. Most of this material has been submitted in previous reports, but it is believed that this summary brings together all information essential to an understanding of the cold-hardiness of the mountain pine beetle in the northern Rocky Mountain region.

Some time we would like to work upon the cold-hardiness of mountain pine beetle parasites and predators. It is possible that abnormal mortality of these beneficial insects as a result of low or unseasonal temperatures may have a direct bearing upon the fluctuations of this important forest insect.

Copies of the report have been sent to the Portland, California, and Fort Collins Laboratories, and a copy routed through the eastern laboratories.

Your comments will be appreciated.

James C. Evenden

Enclosure

SUMMARY REPORT OF LOW TEMPERATURE STUDIES
1936 - 1942

INDEX

SUMMARY REPORT OF LOW TEMPERATURE STUDIES
1936 - 1942

by James C. Evenden, Senior Entomologist [1]

INTRODUCTION

The usual mortality in overwintering bark beetle broods is often as high as 90 percent or more. A slight departure from this high mortality may result in a marked change in the population of these destructive forest enemies. Instances of abnormally high brood mortalities have reduced the seriousness of specific bark beetle outbreaks and eliminated the need for previously planned control operations. Extreme low temperatures have been offered in explanation of these beneficial phenomena, and in some instances the association seemed unquestionable. However, the oddurrence of such conditions during winters of normal temperatures prevented the full acceptance of this explanation and evidenced the need for the consideration of other factors. The potentials of these unusually high bark beetle mortalities carried sufficient economic value to warrant a thorough study of the vulnerability of mountain pine beetle larvae to low temperatures. This project was started in November 1936 and continued each winter until March 1942. This prolonged period resulted from a necessary change in the technique of operation and the occurrence of two mild winters when existing temperatures did not develop sufficient larval cold-hardiness to check results previously obtained. This manuscript summarizes the work that has been done and draws conclusions. Although it can not be said that this project has been fully completed, it is believed that its practical demands have been answered.

LABORATORY PROCEDURES

In conducting this study, techniques of operation previously developed at the Forest Insect Laboratory, Berkeley, California, were followed as closely as possible. Bark beetle larvae were removed from infested logs and placed in specially prepared petri dishes. Although this procedure was simple in operation, it was not without objections. These objections included the disturbance from natural hibernating conditions, the danger of injury to the larvae, as well as the task of determining the mortality associated with each exposure. An effort was made to surmount these objections, but it is not known how successful it was. The most serious of these problems was the difficulty of determining the mortality associated with each experimental exposure, as the freezing of the larvae was no indication of immediate death.

Larvae for each series of tests were obtained from short infested logs that had been stored some 18 miles east of Coeur d'Alene. This cache

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1/ The writer is indebted to Mr. T. T. Terrell, who gave valuable assistance in the planning and conducting of this project.

was located in the mountains under conditions similar to where the logs were cut, to permit as near a normal development of cold-hardiness under actual winter conditions as possible. As required, the logs were transported to Coeur d'Alene, the larvae removed as quickly as possible under outside air temperatures, and then stored in petri dishes at below-activity temperatures. The petri dishes in which the larvae were stored contained a layer of blackened paraffin in which 50 small cells for individual larvae had been constructed. After an exposure to a specific temperature the dishes of larvae were immediately placed in iced containers and allowed to warm as the ice melted. Proper moisture condition within the dish was maintained with a small bit of saturated blotting paper.

Examinations of the treated larvae were made at 24-hour intervals until no further mortality data were available. Although most instances of survival and death were quite evident, border-line cases of mortality presented a difficult problem. When this difficulty arose it was thought that if the treated larvae fed upon fresh phloem, it would be sufficient evidence of recovery. Although this plan offered some data, they were not positive, as a number of severely injured larvae did some feeding while others which seemed to have fully recovered failed to feed under the conditions of the experiment. Although still questionable, the procedure finally adopted was to consider all larvae that responded to a slight shaking of the petri dish with an active body movement as having survived the exposure. When stimulated under microscopic observation, many larvae responded with a slight body pulsation and, in some instances, after 48 to 60 hours seemed to effect a complete recovery. Larvae judged as having resisted the effects of an exposure were subjected to further tests to determine the permanency of this condition. Many of them feed upon fresh phloem, while some prepupal larvae carried through their development to well-formed pupae.

Although comparative data were obtained, the difficulty of determining mortality, the removal of the larvae from their normal place of hibernation, and the possible disturbance of natural processes of hardening or cold-hardiness development made the preceding technique of operation rather unsatisfactory. This dissatisfaction prompted the use of a short infested log, in lieu of the larvae in petri dishes, for each exposure. Although this plan entailed more labor, more material and time, it was considered as providing data which could be accepted by the operator with as little question as possible.

COLD-HARDINESS OF MOUNTAIN PINE BEETLE LARVAE

A large number of experiments were conducted in determining the resistance of mountain pine beetle larvae to low temperatures. As many of these were necessary in the development of operative techniques and in the checking of results, they will not be included in this summary. During the first winter's work it was only possible to record the cold-hardiness of midwinter and early spring larvae. A brief description and the results of these two experiments follow:

Resistance of Midwinter Larvae of the
Mountain Pine Beetle to Specific Temperatures

Hosts - Lodgepole pine - Ashton, Idaho, and Mount Washburn, Yellowstone Park

White pine - Coeur d'Alene, Idaho

Date - January 26, 1938.

Description of experiment - Larvae were taken from two hosts representing three separate localities, and exposed for 2 hours and 15 minutes to set temperatures varying from 0 to -30° F. Before and after exposure the larvae were treated as previously described. The results of this experiment are shown in the following tabulation:

Table 1 - Cold-hardiness of <u>midwinter</u> mountain pine beetle larvae from white pine and lodgepole pine

2 hour-15 minute exposure	Hosts	Mortality 48 hours after exposure - dead larvae only	Mortality 48 hours after exposure. Includes dead and quiescent larvae.
0°F	Lodgepole - Ashton, Idaho	2%	14%
	Lodgepole - Yellowstone Park	4%	14%
	White pine - Coeur d'Alene, Ida.	2%	2%
- 5°F	Lodgepole - Ashton	0	12%
	Lodgepole - Yellowstone	0	8%
	White pine - Coeur d'Alene	0	2%
-10°F	Lodgepole - Ashton	0	12%
	Lodgepole - Yellowstone	4%	10%
	White pine - Coeur d'Alene	8%	8%
-15°F	Lodgepole - Ashton	2%	10%
	Lodgepole - Yellowstone	0	6%
	White pine - Coeur d'Alene	0	4%
-20°F	Lodgepole - Ashton	0	16%
	Lodgepole - Yellowstone	0	6%
	White pine - Coeur d'Alene	0	2%
-22.5°F	Lodgepole - Ashton	0	18%
	Lodgepole - Yellowstone	6%	14%
	White pine - Coeur d'Alene	0	4%
-25°F	Lodgepole - Ashton	2%	12%
	Lodgepole - Yellowstone	0	22%
	White pine - Coeur d'Alene	0	10%
-27.5°F	Lodgepole - Ashton	4%	4%
	Lodgepole - Yellowstone	6%	6%
	White pine - Coeur d'Alene	6%	8%
-30°F	Lodgepole - Ashton	0	4%
	Lodgepole - Yellowstone	4%	8%
	White pine - Coeur d'Alene	6%	10%
Checks	Lodgepole - Ashton	0	22%
	Lodgepole - Yellowstone	0	2%
	White pine - Coeur d'Alene	0	0

Although the insects used in this experiment represented two hosts from three distinct localities, all logs had been assembled in early fall and stored in the same cache during the winter. This may have resulted in an equalization of the larval resistance shown by these data, as an increased cold-hardiness could be expected from the colder areas at Ashton, Idaho, and Mount Washburn, Yellowstone Park. It is recognized that if lower temperatures had been available this area difference in resistance might have manifested itself. The light mortality distributed throughout all exposures can only be explained by the presence of a few

non-resistant larvae. The conclusion to be drawn from these data is that an exposure of 2 hours and 15 minutes to a temperature of -30° F. was not lethal to these larvae.

Resistance of Early Spring Larvae of the Mountain Pine Beetle to Specific Temperatures

Hosts - Lodgepole pine - Ashton, Idaho; Mount Washburn, Yellowstone Park

Whitebark pine - Mount Washburn, Yellowstone Park

White pine - Coeur d'Alene, Idaho

Date - May 11, 1937.

Description of experiment - Larvae were taken from each of the three hosts, which represented four localities, and exposed to varying temperatures for 2 hours and 15 minutes. The treatment of the larvae prior to exposure varied from previously described procedures, as after being removed from the logs they were stored at outside temperatures. As larval activity had started in all logs, with a few newly formed pupae and new adults, storage at below-activity temperatures could have developed an abnormal resistance.

Table 2 - Temperatures lethal to mountain pine beetle larvae subsequent to the start of spring temperatures

Exposure for 2 hours and 15 minutes	Lodgepole Ashton, Idaho		Lodgepole Yellowstone Park		Whitebark pine Yellowstone Park		Western white pine Coeur d'Alene, Idaho	
	Alive	Dead	Alive	Dead	Alive	Dead	Alive	Dead
+ 15°F	98%	2%	98%	2%	100%	0	96%	4%
+ 10°F	84%	16%	94%	6%	12%	88%	26%	74%
+ 5°F	50%	50%	52%	48%	0	100%	18%	82%
0	10%	90%	8%	92%	-	-	-	100%
- 5°F	-	100%	-	100%	-	-	-	-
Check	100%	-	100%	-	100%	-	100%	-

These data show that when mountain pine beetle larvae are exposed to activity temperatures their cold-hardiness is broken, with a temperature of -5° F. being lethal to the insects from all hosts and localities. These data were checked by the following experiment: Three dishes of 50 larvae from each host were removed from infested logs and immediately

exposed to a constant temperature of -2.5° F., with one dish being removed at the end of each 20-minute period. The results of this test again show the effects of activity temperatures upon larvae which had previously resisted exposures to -30° F.:

Table 3

Exposure at -2.5°F	Lodgepole Ashton, Idaho		Lodgepole Yellowstone Park		Whitebark pine Yellowstone Park		Western white pine Coeur d'Alene, Idaho	
	Alive	Dead	Alive	Dead	Alive	Dead	Alive	Dead
20 minutes	100%	0	94%	6%	100%	0	94%	6%
40 minutes	48%	52%	24%	76%	10%	90%	8%	92%
60 minutes	10%	90%	2%	98%	2%	98%	0	100%

The slight mortality that occurred at the end of the 20-minute period could have resulted from injured larvae, as none of them were frozen when removed, or it could have been the start of the severe mortality that occurred at 40 minutes, as all larvae were frozen at that time. The conclusion drawn from these experiments was that the cold-hardiness of mountain pine beetle larvae was seasonal in character and followed a period of tempering or preparatory temperatures.

Seasonal Development of Cold-hardiness
of Mountain Pine Beetle Larvae

As previous experiments seemed to demonstrate that cold-hardinese was a matter of preparedness, experiments were conducted to show the seasonal development and subsequent reduction in the resistance of mountain pine beetle larvae to extreme low temperatures.

Host - Western white pine - Coeur d'Alene, Idaho

Date - September 1937 - May 1938

Description of experiment - To obtain these data it was necessary to conduct tests at 15-day intervals from September 18, 1937, to June 1, 1938. This plan was followed; however, during December, January, and February larval resistance was greater than the lowest temperature attainable with available equipment. The same technique of using exposed larvae in petri dishes was employed as with previous experiments. The range of temperatures was planned to include both extremes, although it was recognized that lethal temperatures could not be obtained if the larval resistance was equal to that of the previous season.

The results of these experiments are presented in charts I and II. In chart I the results of each test are shown, while in chart II curves of

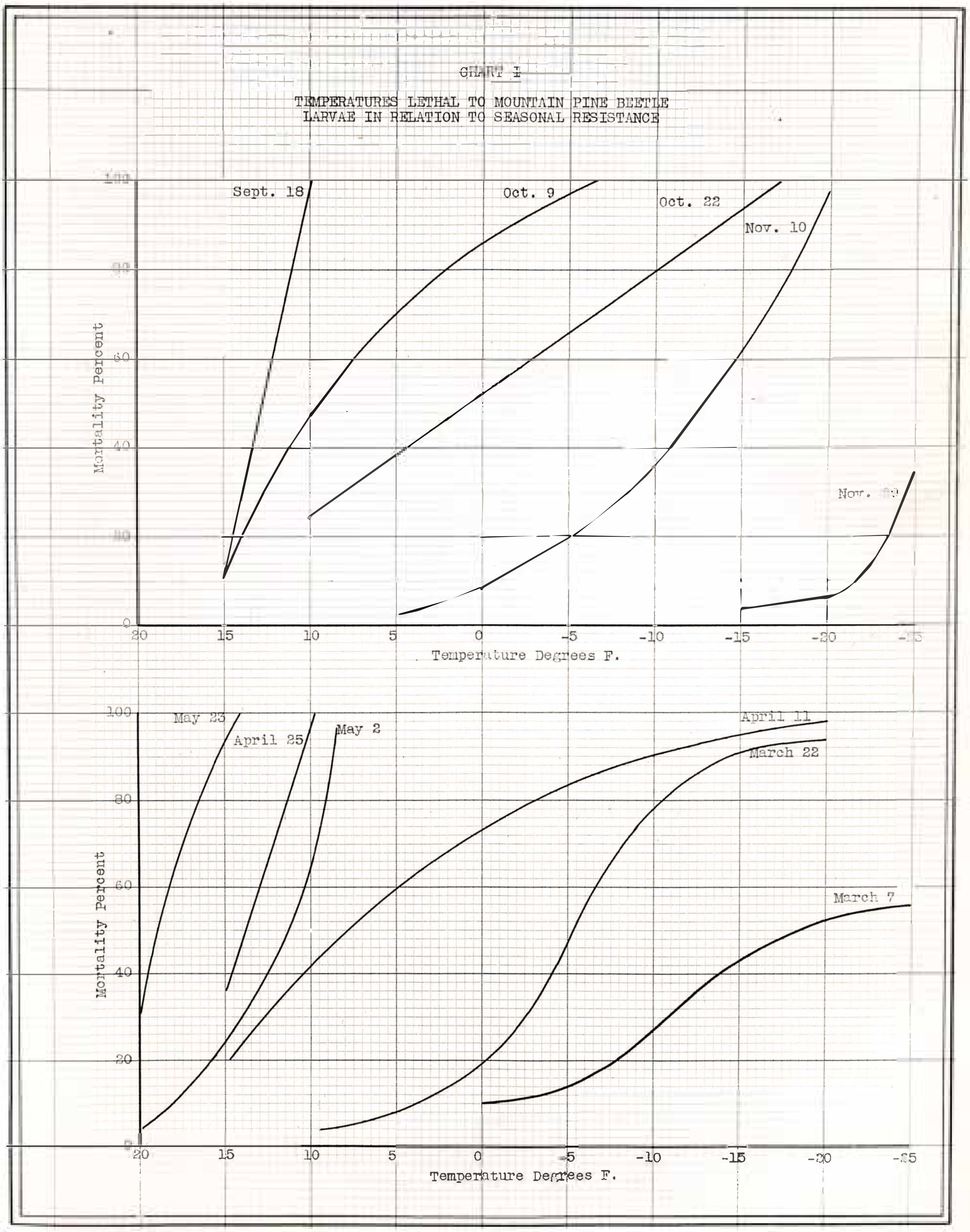
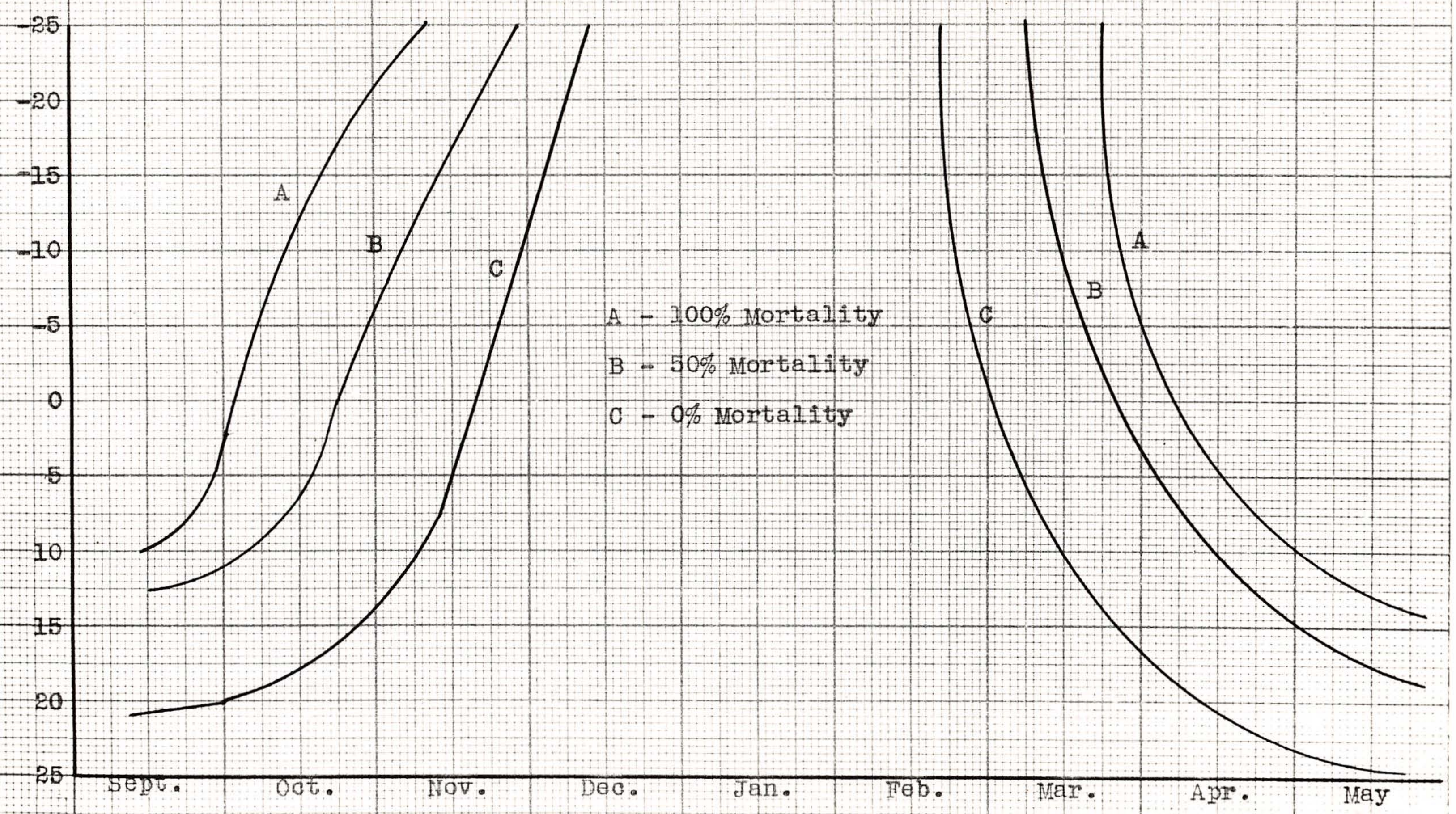


CHART I
TEMPERATURES LETHAL TO MOUNTAIN PINE BEETLE
LARVAE IN RELATION TO SEASONAL RESISTANCE
Sept. 18
Oct. 9
Oct. 22
Nov. 10
Mortality Percent
100
80
60
40
20
0
20
15
10
5
0
-5
-10
-15
-20
-25
Temperature Degrees F.
May 23
April 25
May 2
April 11
March 22
March 7
Mortality Percent
Temperature Degrees F.

CHART II

SEASONAL CHANGES IN THE RESISTANCE OF MOUNTAIN PINE BEETLE LARVAE TO LOW TEMPERATURES



no mortality, 50 percent mortality and 100 percent mortality are presented. It is regretted that the maximum resistance could not be determined; however, it was apparent that the larvae again withstood exposures of 2 hours and 15 minutes to temperatures as low as -30° F.

This series of tests was repeated the following winter (1938-39) and an effort made to overcome the objection of non-lethal cabinet temperatures and obtain lower temperatures by placing dry ice in the cabinet. This plan proved fairly satisfactory, but as the temperatures obtained were below the functional limits of the thermostatic control, the desired temperatures were obtained by placing an electric light in the cabinet. A constant checking of the temperature within the cabinet with a potentiometer and by switching the light on and off maintained the desired temperature within a range of ± 1° F. Although each test was planned to include the range of lethal temperatures, in a few instances errors were made in determining the point at which larval mortality started. Data obtained from this series of tests are shown in chart III on the following page.

From chart III it will be seen that there was a steady increase of larval resistance from August 1938 to January and February 1939. This resistance was broken in March by warm weather, and by May 1 it had decreased to a degree comparable to that of the previous August.

The data recorded for October 10 are sufficiently out of line to warrant explanation. The larvae for this test were collected in the field and were from the same infested log as those used in the September 24 and October 24 experiments. On October 10 the weather was moderate, with a warm rain throughout the day. Although the larvae were active when collected, it is possible that whatever cold-hardiness existed at that time was reduced somewhat while the larvae were transported to the laboratory. This instance of reduced resistance illustrates the close adherence of cold-hardiness to changes in temperatures.

Data obtained on January 12 illustrate the extreme temperatures used during these tests. Larvae which recovered from the -61°F exposure did some feeding on fresh white pine phloem, but soon became inactive and died. Larvae from the -78°F exposure lived until January 22, but did not feeding. This test of extreme temperatures was carried further than shown on the chart, and larvae exposed to -116°F. Although 68 of these larvae recovered and became active, they all died before January 22. These data indicate only a temporary recovery of mountain pine beetle larvae from freezing. Regardless of its duration such temporary recovery is only a measure of cold-hardiness for the purposes of this experiment and does not depict larval resistance under normal field and hibernating conditions.

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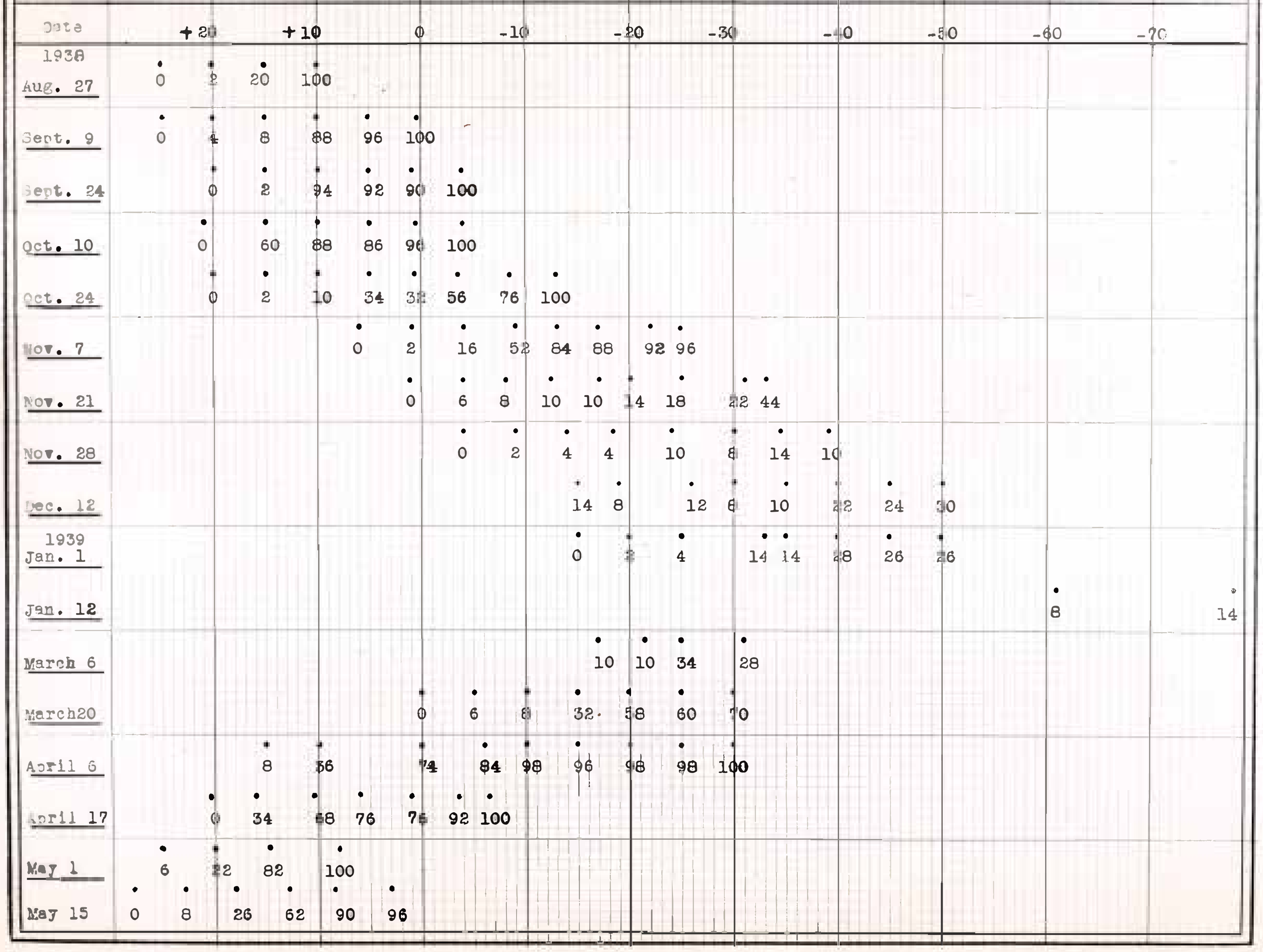
CHART III
SEASONAL RESISTANCE OF MOUNTAIN PINE BEETLE LARVAE TO LOW TEMPERATURES
WHITE PINE
Larval Mortality Shown in Percent



A range of temperatures in relation to the mortality of mountain pine beetle larvae from white pine for the winter of 1938-39 is shown in chart IV. The curve depicting the temperature at which no mortality occurred is a smoothed curve plotted on the highest temperature points.

Monthly minimum air temperatures at Coeur d'Alene, Idaho, are compared to the trend of these curves to show the relation of cold-hardiness development to winter temperatures. Although the air temperature records do not depict conditions at the log cache, they do serve to show the relationship.

A sharp increase in larval resistance is shown in the 100 percent mortality curve for May 1939. This increase resulted from the testing of pupae and prepupal larvae.

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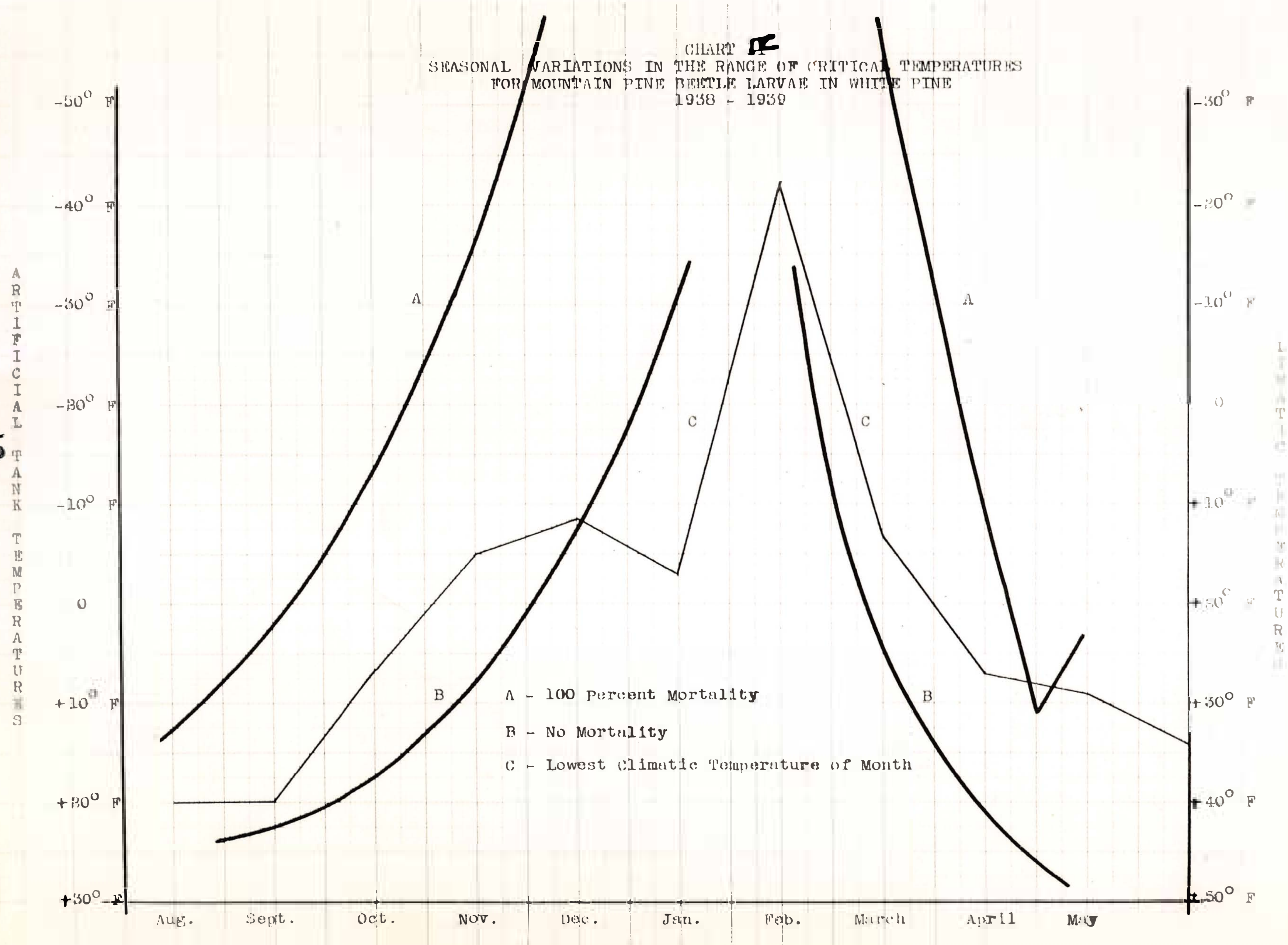


CHART II
SEASONAL VARIATIONS IN THE RANGE OF CRITICAL TEMPERATURES
FOR MOUNTAIN PINE BEETLE LARVAE IN WHITE PINE
1938 - 1939
ARTIFICIAL TANK TEMPERATURES
-50° F
-40° F
-30° F
-20° F
-10° F
0
+10° F
+20° F
+30° F
-30° F
-20° F
-10° F
0
+10° F
+20° F
+30° F
+40° F
+50° F
CLIMATIC TEMPERATURES
A
A
C
C
B
B
A - 100 Percent Mortality
B - No Mortality
C - Lowest Climatic Temperature of Month
Aug.
Sept.
Oct.
Nov.
Dec.
Jan.
Feb.
March
April
May

To avoid confusion in the interpretation of the results obtained as well as to provide more positive mortality data, subsequent experiments were conducted under as near natural conditions as possible. Short infested logs from 10 to 16 inches in diameter and 24 inches in length were used in lieu of larvae in petri dishes. Thermocouples were inserted under the bark and the subcortical temperature of the log read with the aid of a potentiometer. The temperature of the cabinet was set at outside conditions for the reception of the log, and then gradually lowered until the desired exposure had been obtained. Only a few minutes were required to place the logs in the cabinet, which eliminated possible disturbances from normal conditions. After the logs were removed from the cabinet they were allowed to stand at room temperatures from 10 to 14 days before being examined. This procedure eliminated all borderline cases of questionable mortality, as all dead larvae were blackened or swollen, while those that had withstood the exposure were feeding actively, with many newly formed pupae and new adults. Each series of tests was protected with check logs to show the brood status at the time of exposure and to eliminate any question as to the cause of mortality. Although this procedure required more time for each test, as well as a larger stock of infested logs, accurate mortality data were obtained under near normal conditions as possible in the laboratory.

The first two seasons that these logs were used the cold-hardiness of the larvae was not comparable to that of the previous seasons. Mild winters are believed to be responsible for this condition, for although the larvae withstood existing field temperatures they did not show the same degree of resistance against laboratory temperatures.

During the winter of 1939-40, a 99+% mortality was obtained in all tests at -30°F. In February 1940 brood mortalities of 98 and 78 percent followed exposures to temperatures of -25 and -20°F. This lack of resistance was still further shown in March, when mortalities of 97 and 52 percent were obtained from exposures to -21 and -15°F. In April a 97% mortality followed an exposure to 0° F.

The following winter (1940-41) there was a slightly higher degree of resistance, which is believed to have resulted from the occurrence of lower temperatures in November 1940. In this series of tests the following data were obtained:

Table 4 - Mortality of mountain pine beetle larvae when exposed to a lowering temperature to produce desired subcortical conditions

1940-41

Date	Mortality Percent	Limits of exposure: Subcortical	Limits of exposure: Cabinet
Dec. 2-6, 1940	0	38 to -10°F	42 to -16°F
	57	43 to -15°F	48 to -21°F
Jan. 6-12, 1941	0	32 to -20°F	32 to -23.8°F
	25	32 to -25°F	32 to -26°F
	31	32 to -30°F	32 to -30.15°F
Feb. 3-7, 1941	75	35 to -20°F	31.1 to -23.85°F
	88	32 to -25°F	31.1 to -26.9°F
	95	32 to -30°F	28.6 to -30.15°F
	94	31.6 to -35°F	28.6 to -35°F
Feb. 13-19, 1941	21	38.5 to -20°F	41.3 to -23.35°F
	61	36.2 to -25°F	41.3 to -28.5°F
	100	28.6 to -30°F	25.8 to -30°F
Mar. 17-20, 1941	98	44.3 to -10°F	46 to -18.3°F
	100	45 to -15°F	46 to -23.35°F

The writer frankly admits poor guessing in determining the critical temperatures for these tests, which detracts somewhat from their value.

February tests of this experiment were checked with exposed larvae in petri dishes. Although the data obtained indicate that exposed larvae have a higher degree of resistance, this is not significant due to the difficulty of determining actual mortality.

These experimental tests were repeated again during the winter of 1941-42. Although some cold weather was experienced for a day or two during the later part of December, the early fall temperatures were not considered as being sufficiently low to develop a degree of cold-hardiness in mountain pine beetle larvae comparable to that which existed during the three winters from 1936 to 1939. Although complete lethal temperatures were not obtained in all tests, as only the minimum temperature limit of the cabinet was used, it is believed that these data with those already made available satisfy the practical demands of the problem. The results of the 1941-42 tests are shown in the following tabulation:

Table 5 - Brood mortality in white pine logs exposed to lowering temperatures to produce different subcortical temperatures

1941-42

Date	Mortality Percent	Limits of exposure Subcortical	Cabinet
Dec. 2, 1941	0	43 to -5°F	33.4 to -13.7°F
	3.7	43 to -10°F	33.4 to -16.8°F
Dec. 4, 1941	12.8	35.3 to -15°F	39 to -22.2°F
	31.6	35.3 to -20°F	39 to -25°F
Dec. 6, 1941	76.0	32.9 to -25°F	35.3 to -28.50°F
	100.0	32.9 to -30°F	35.3 to -33.15°F
Dec. 29, 1942	0	29.7 to -25°F	22.5 to -29.6°F
	76	29.7 to -30°F	22.5 to -32.65°F
Jan. 7, 1942	9	29.7 to -25°F	22.5 to -29.6°F
	48	31.1 to -30°F	22.5 to -30.1°F
Feb. 6, 1942	0	33.9 to -22.50°F	30.2 to -24.4°F
Feb. 2, 1942	28.6	32.0 to -25.00°F	32 to -25.8°F
Feb. 6, 1942	91.2	33.9 to -27.50°F	30.2 to -30.1°F
Feb. 2, 1942	96.8	32.0 to -32.50°F	32 to -32.5°F
Mar. 2, 1942	0	32 to -20.00°F	26 to -24.4°F
	25	32 to -25.00°F	26 to -29.1°F
Mar. 4, 1942	100	31.6 to -28.00°F	26.6 to -30.1°F
Apr. 10, 1942	100	33.0 to -3.75°F	32.0 to -15°F

Although the preceding data do not show the maximum limits of cold-hardiness, they do demonstrate that the resistence of the larvae was more than sufficient to withstand seasonal temperatures. During the winter of 1941-42 the minimum temperature (Coeur d'Alene City record, which was perhaps a few degrees higher than at the log cache) was but -11°F. The cold-hardiness of the larvae as shown by this experiment indicated an ample margin of safety.

These data stress again the variation in the resistance of individual larvae, as it is evident that at no time do all larvae, even from the same portion of an infested log, acquire the same degree of cold-hardiness. Although the stage of development is a comparative measure of larval resistance to low temperatures, with prepupal larvae the most resistant, the variation of cold-hardiness in individual larvae is undoubtedly influenced by other factors as well. The development and reduction of larval resistance to low temperatures is rather clearly illustrated by the preceding data.

MISCELLANEOUS EXPERIMENTAL TESTS

An evaluation of the resistance of mountain pine beetle larvae to low temperatures involved the answers to a number of questions. Insulation of bark, duration of exposures, establishment and breaking of larval resistance, as well as different operative techniques, were associated with this problem. The answers to some of these essential questions follow.

Insulation or Protection Afforded Bark Beetle Larvae by Normal Covering of Bark

Before the lethal effects of low air temperatures could be determined it was necessary to know something of the lag which could be expected between air and subcortical temperatures. As it was necessary also to know if subcortical temperatures were affected by different-sized logs, the following experiments were conducted:

Objective - To determine the lag between air, subcortical, and log-center temperatures.

Host - Lodgepole pine. Mountain pine beetle larvae.

Date - March 11, 1937.

Description of experiment - Both ends of an infested log 13 inches in diameter and 24 inches in length were covered with discs of corrugated cardboard, and the edges sealed with paraffin. (Although subsequently found to be unnecessary, this precaution was taken to guard against the possibility of abnormal subcortical temperatures resulting from the exposed bark ends.) A thermocouple was buried 6-1/2 inches deep to the center of the log midway from each end, and others placed beneath the bark at 2, 4, 6, and 12 inches from the ends of the log. The 2-, 4-, and 6-inch installations were used to check the possibility of abnormal exposure from the log ends. Thermocouples were inserted in a 3/32-inch hole, which was then packed solid with cotton and the surface sealed with paraffin. (Sealing with paraffin later found unnecessary.) Holes for the subcortical thermocouples were 1-1/2 inches long and drilled at an angle to follow the curve of the log. To eliminate the effects of winter temperatures the log was held in the laboratory for several days prior to the test. After being exposed to a lowering temperature to a point when all thermocouples had registered the same for several hours, the compressor was stopped and the cabinet allowed to warm slowly to the same temperature at which the experiment was started. Results are shown in chart V.

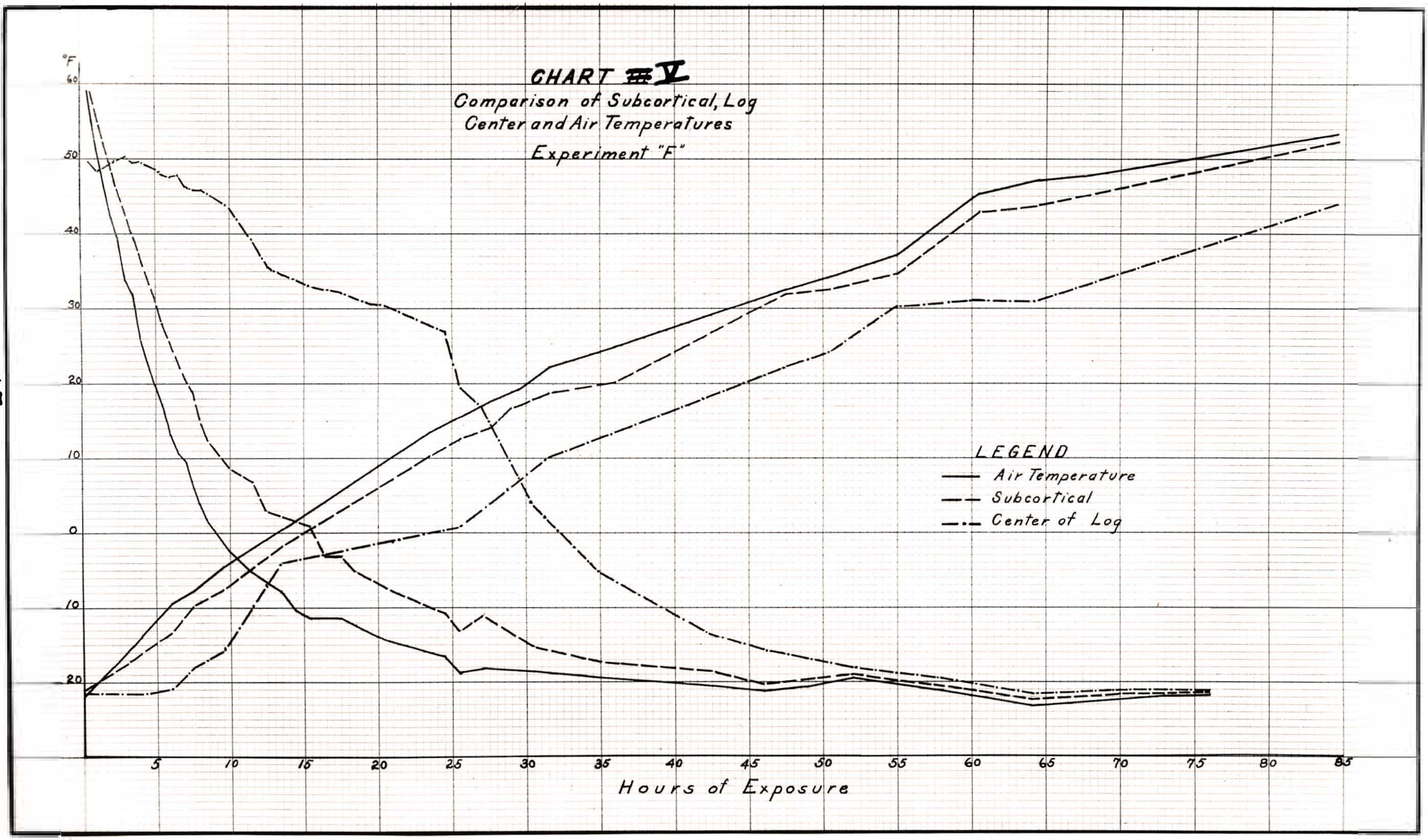


CHART ~~III~~ V
Comparison of Subcortical, Log Center and Air Temperatures
Experiment "F"
°F
60
50
40
30
20
10
0
10
20
5
10
15
20
25
30
35
40
45
50
55
60
65
70
75
80
85
Hours of Exposure
LEGEND
Air Temperature
Subcortical
Center of Log

These data illustrate the lag between the subcortical and air temperatures. The course these curves have followed is as one would have anticipated. Approximately five hours were required for the rapidly decreasing air temperature to affect the center of the log. As the air temperature of the cabinet leveled off, the subcortical as well as the log center dropped rapidly to the same level. As the temperature within the cabinet increased, subcortical and log-center temperatures followed quite closely, although five or six hours were again required before the center of the log was influenced by this condition. It will be seen that the log temperatures respond more quickly to an increasing than they do to a decreasing air temperature.

As data obtained from thermocouples at the 2-, 4-, and 6-inch placements from the end of the log showed practically no difference from the 12-inch location, it is concluded that no undue exposure occurred. These data are not shown, as the slight variation did not permit plotting.

As a check upon these results a slightly different experimental set-up was conducted. Thermocouples were placed under the bark and at depths of 1, 2, 3, and 6 inches in the wood. The log was held at room temperatures for several days and then exposed to a lowering temperature which started at 69.4°F and ended at -20.3°F. At the minimum temperature attainable the compressor was stopped and the cabinet warmed to room temperatures. The results of this check test are shown in chart VI on the next page.

It will be seen that this check test provided the same results as obtained with the original test. The dip in the curves showing the increasing temperatures illustrates the rapidity at which subcortical and log temperatures follow changes in air conditions. As the log was warming the compressor was started and the temperature of the cabinet reduced from 28.5 to 22.8°F. The results of this action are self-evident.

These data show the lag between air and subcortical temperatures is dependent somewhat upon the speed at which the air temperature changes. As normal air temperatures change more rapidly than that under which these laboratory experiments were conducted, the natural lag of subcortical temperatures to a decreasing air temperature would be greater than indicated by the preceding data. This lag of subcortical temperatures with their quick response to increasing air temperatures would permit the occurrence of lethal air conditions in the field, with no resulting ill effects upon overwintering bark beetle broods.

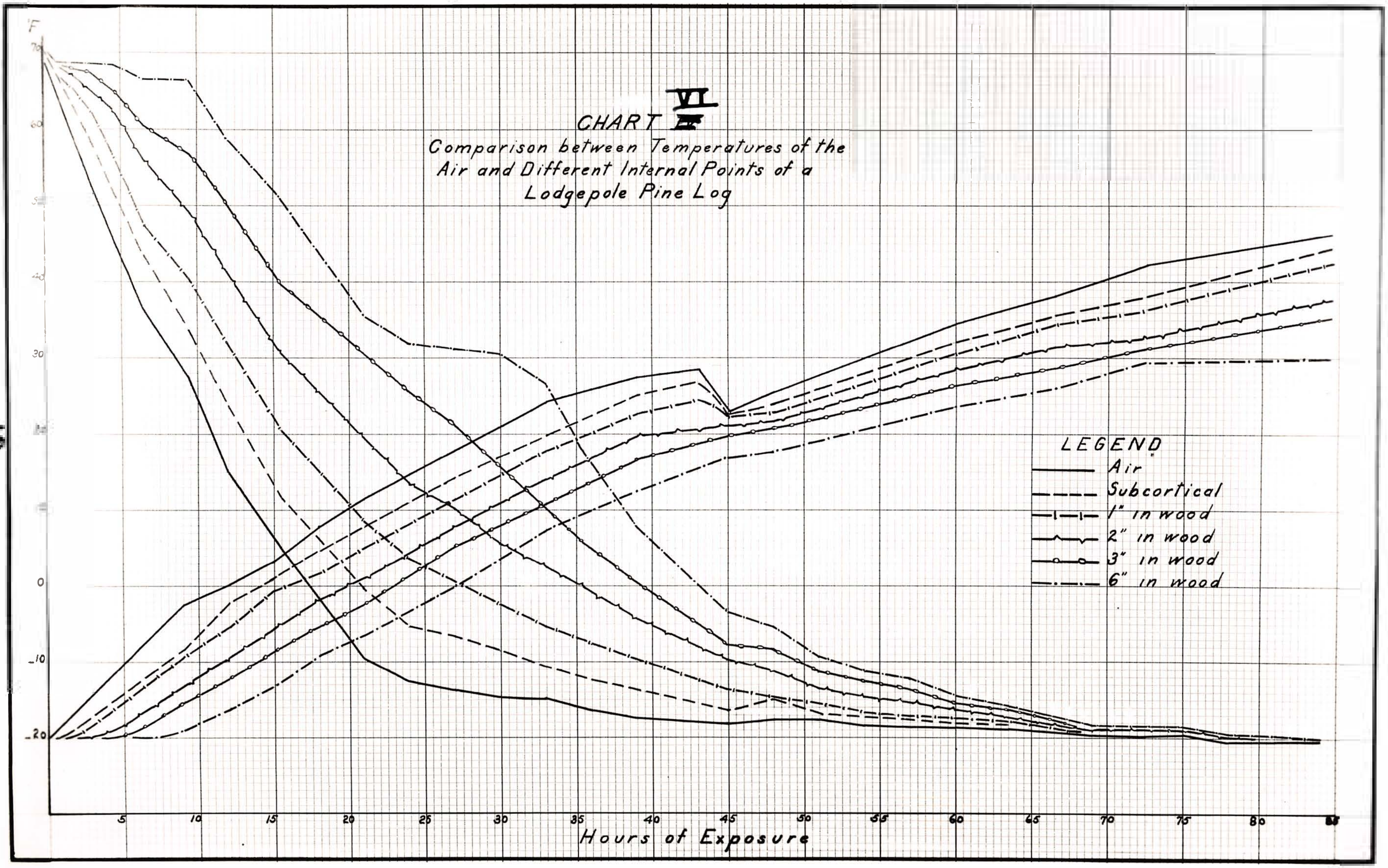


CHART VI
Comparison between Temperatures of the Air and Different Internal Points of a Lodgepole Pine Log

Log Diameters and Subcortical Temperatures

As it was thought that large trees or logs might possess some stored heat that would influence the relation between subcortical and air temperatures, the following experiment was conducted:

Objective - To determine the influence of different tree diameters upon subcortical temperatures.

Host - Lodgepole pine

Date - April 22, 1937.

Description of experiment - Three logs 24 inches in length and 7, 11, and 14 inches in diameter were held under laboratory conditions for several days and then exposed to a temperature which started at 60.7°F and ended at -22.10°F. Thermocouples were placed under the bark, 1 inch in the wood, and in the center of each log. Data relative to bark thickness and wood texture were taken for each log.

Table 6

Log	Diameter	Bark thickness	Annual Rings		
			Outside inch	Outside 2 inches	Outside 3 inches
1	7"	.10"	41	76	93
2	11"	.20"	15	27	34
3	14"	.15"	16	27	36

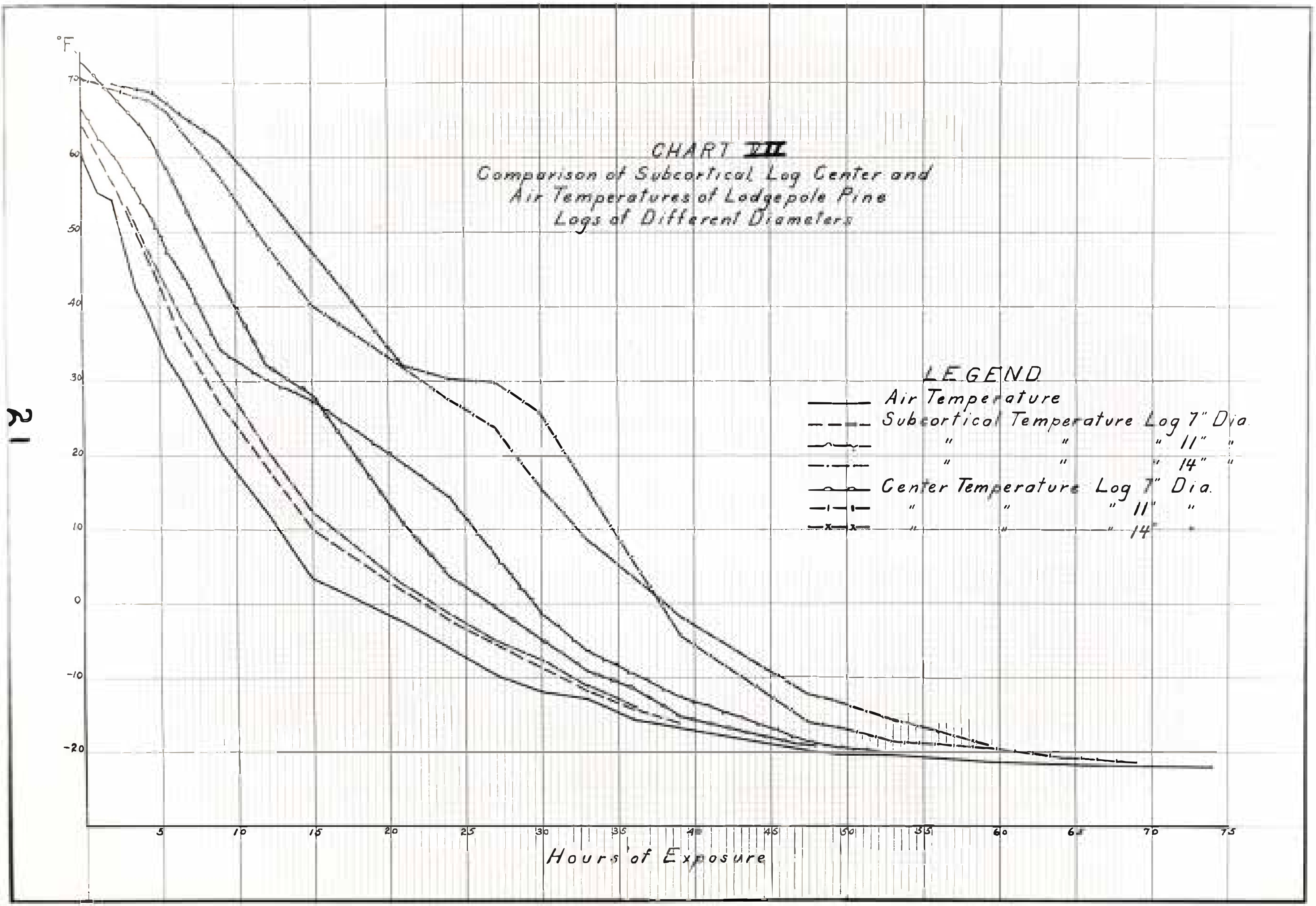


CHART VII
Comparison of Subcortical, Log Center and
Air Temperatures of Lodgepole Pine
Logs of Different Diameters
LEGEND
Air Temperature
Subcortical Temperature Log 7" Dia.
" " " 11" "
" " " 14" "
Center Temperature Log 7" Dia.
" " " 11" "
" " " 14" "
°F.
70
60
50
40
30
20
10
0
-10
-20
5
10
15
20
25
30
35
40
45
50
55
60
65
70
75
Hours of Exposure

Although these data fail to show any relation between log diameters and the lag between subcortical and air temperatures, they do show that bark thickness is a governing factor. It will be noted that the subcortical temperature of the 14-inch logs adhered closely to that of the 7" log, which would seem to eliminate the question of diameter. The slowest growing tree (7" log) showed the closest adherence to air temperature; however, as the bark on this log was quite thin, it is assumed that this is a more significant factor than the rate of growth. These data show, as would be expected, a positive relation between bark thickness and subcortical and log center temperatures. The drop of the log center temperature of the 7" log below the subcortical temperature of the 11" log might seem to be somewhat confusing. However, this occurrence would be expected, as the center temperature of the small, thin-barked log would obviously soon drop below the subcortical temperature of the larger, thick-barked 11" log. This is also true of the crossed log center temperatures of the 11" and 14" logs, which again can best be explained by the protection afforded by the different bark thickness.

Length of Exposure

Weather records show the occurrence of low temperatures for two or more days, with little difference between maximum and minimum readings. The following series of experiments was planned to check the possibility of prolonged exposures resulting in greater bark beetle mortalities. In connection with all previous tests an exposure of 2 hours and 15 minutes was adopted as a standard procedure, to provide a 2-hour exposure for larvae in covered petri dishes. The following experiments were conducted to determine if the length of exposure influenced the resulting bark beetle mortality.

I

Objective - To determine if difference existed in the effects of 1¼- and 2¼-hour exposures.

Insect - Mountain pine beetle in white pine.

Date - March 25, 1938

Description of experiment - Groups of 250 and 200 larvae in petri dishes were exposed to a temperature of -6°F for 1¼ and 2¼ hours respectively. The data obtained are shown in the following tabulation. Normal resistance of mountain pine beetle larvae on March 22, 1938, was 45% at -6°F.

Table 7 - Larval mortality resulting from exposures of
1¼ and 2¼ hours

Dish	Temp. F.	Mortality	Remarks
		Exposure 1 hour 15 minutes	
1	-6	36%	Average mortality for this group of
2	"	46%	250 larvae was 39.6%. Mortality in
3	"	42%	the individual dishes was sufficiently
4	"	30%	close as to indicate representative
5	"	44%	samples.
		Exposure 2 hours 15 minutes	
6	-6	34%	Average mortality for this group of
7	"	38%	200 larvae was 33% or 6% lower than
8	"	26%	the shorter exposure. This difference
9	"	34%	can not be considered as significant
			of the two treatments.

Although these data show no difference in brood mortality for the different treatments, they are not entirely significant, as such a difference could have existed had more lethal temperatures been used.

II

Objective - Relation of accumulative low temperatures to the mortality of mountain pine beetle larvae.

Hosts - Lodgepole pine - Ashton, Idaho

Lodgepole pine - Yellowstone Park

Date - February 1-11, 1937.

Description of experiment - Petri dishes of larvae from each host were exposed to a set temperature, with one dish being removed at the end of each 2-hour period. The same technique of treatment before and after exposure was used with all tests of petri dish larvae. Results obtained are given in the following tabulation:

Table 8 - Effects of accumulative temperatures on mountain pine beetle larvae from lodgepole pine

Set temperature	Percent of mortality 48 hours after exposure: 2 hours	4 hours	6 hours	8 hours	10 hours	Check larvae
	Lodgepole pine from Ashton, Idaho					
0°F	0	0	2	2	0	0
-5°F	4	4	6	4	2	0
-10°F	8	6	4	2	6	2
-15°F	10	12	2	4	10	6
-20°F	4	2	4	4	8	0
-25°F	8	6	10	12	2	4
	Lodgepole pine from Yellowstone Park					
0°F	2	2	4	2	4	4
-5°F	14	24	18	10	4	0
-10°F	Larvae were not available for this test					
-15°F	2	6	10	8	2	6
-20°F	2	2	2	0	2	6
-25°F	10	8	4	4	8	6

This experiment also fails to indicate any difference in the effects of 2- and 10-hour exposures, although perhaps more lethal temperatures would have demonstrated such a difference. The mortality of nonresistant larvae is quite pronounced in these results. To carry the thought of accumulative exposures to an extreme, the following experiment was conducted:

Objective - To determine the effects of prolonged exposures to low temperatures upon the mortality of mountain pine beetle larvae.

Insect and host - Mountain pine beetle in white pine

Date - December 27, 1937, and January 7, 1938.

Description of experiment - A number of petri dishes of larvae (50 each) were all placed in the low-temperature cabinet at one time and exposed to a set temperature, with one dish being removed after a definite period of time. The results obtained are shown in the following tabulation:

Table 9 - Mortality of mountain pine beetle larvae following prolonged exposures to a temperature of -20°F

Dish	Length of exposure	Percent frozen at end of exposure	Percent mortality
1	2 hrs.	42	2
2	4 "	62	6
3	8 "	80	6
4	16 "	70	4
5	1 day	40	2
6	2 days	96	4
7	3 "	66	2
8	4 "	24	4
9	5 "	34	6
10	6 "	24	8
11	7 "	20	6
12	8 "	--	7.5

Table 10 - Mortality of mountain pine beetle larvae following prolonged exposures to a temperature of -25°F

Dish	Length of exposure	Percent frozen at end of exposure	Percent mortality
1	2 hrs.	22	6
2	4 "	46	0
3	8 "	74	0
4	16 "	84	4
5	1 day	40	6
6	2 days	84	6
7	3 "	92	4
8	4 "	24	8
9	5 "	12	4
10	6 "	26	4
11	7 "	34	12
12	8 "	22	10
13	9 "	32	4
14	10 "	4	8

As all tests indicated no increased mortality for prolonged exposures, it would seem that mortality is associated with a short exposure to a lethal temperature. In these two tests the constant mortality of nonresistant larvae is again quite apparent. No relationship seems to exist between the percent of frozen larvae, which can be easily detected by their color, and the subsequent mortality. However, it has been observed that when all larvae are frozen a high mortality follows.

Reduction or Breaking of Larval
Resistance to Low Temperatures

Although it would seem that the mountain pine beetle develops sufficient cold-hardiness to withstand normal winter temperatures, it was not known if this condition would be broken by the occurrence of abnormal periods of warm weather. It was believed that if this occurred it would be possible for broods of the mountain pine beetle to be made vulnerable to temperatures that would otherwise have caused no mortality. In answer to this possibility the following tests were conducted.

Objective - To determine the effects of abnormally warm temperatures upon established cold-hardiness of bark beetle larvae.

Insect & host - Mountain pine beetle in western white pine.

Date - January 31 and March 8, 1938.

<u>Description of experiment</u> - As complete equipment was not available for these tests, this experiment could only be set up as depicting possibilities. Logs were moved from the cache to the laboratory and exposed for different periods to activity temperatures which varied from 45 to 70°F. After such exposures some logs were returned to the cache to determine if normal resistance would be restored, while the larvae from others were removed for tests of low-temperature resistance. As each groups of logs was brought to the laboratory for exposure to above-activity temperatures, a test of the larvae was made immediately to determine normal resistance.

Test A

Three logs of comparable size and brood conditions were held at laboratory temperatures from January 31 to February 7, 1938. A test of larval resistance on January 31 showed no mortality at -25°F. On February 7 the larvae were removed from one log (A1) and the low-temperature resistance obtained. The other two logs were returned to normal winter temperatures at the log cache. Logs A2 and A3 were returned to the laboratory on February 10 and 21 respectively, and the larval resistance determined. The data obtained are shown in the following tabulation:

Table 11 - Seven day exposure to activity temperatures

Exposure	Log A1 Percent mortality	Log A2 Percent mortality	Log A3 Percent mortality
Normal larval resistance on Feb. 7, 1938. 0% at -25°F			
20°F	6	4	8
15°F	38	38	0
10°F	96	62	44
5°F	94	92	70
0°F	98	100	98
-5°F	100	100	98
-10°F	100	100	100
-15°F	100	100	100

These data show the resistance of the larvae in log A1 to have been reduced from normal cold-hardiness of 0% mortality at -25°F at the time the log was removed from the cache to 100% brood mortality at -15°F. The larvae in log A2 show no recovery from this unseasonal exposure after a return of 3 days to normal winter temperatures at the log cache. Although some increased larval resistance was shown in log A3 after a 14-day exposure to winter temperatures, it was still much below normal resistance on January 31.

Test B

In this test three logs were exposed to laboratory temperatures from February 7 to February 10, 1938. Larvae were removed from log B1 on February 10 and their resistance determined. Logs B2 and B3 were returned to the log cache on the 10th and held until February 21 and March 7 respectively, when they were returned for resistance tests.

Table 12 - Three day exposure to activity temperatures

Exposure	Log B1 Percent mortality	Log B2 Percent mortality	Log B3 Percent mortality
Normal larval resistance on Feb. 7, 1938			0% at -25°F
20°F	0	0	4
15°F	4	0	14
10°F	32	0	26
5°F	40	8	50
0°F	66	20	90
-5°F	96	44	94
-10°F	98	74	90
-15°F	100	90	98

Data in table 12 show the reduced resistance of log B1 after a 3-day exposure, and restored resistance of log B2 after being returned to winter temperatures at the log cache. The somewhat reduced resistance of some larvae in log B2 was no doubt due to the fact that on March 7 the normal resistance had been reduced by warm weather to 56% at -25°F instead of 0% on February 7.

Test C

In this test two infested logs were exposed to laboratory temperatures from February 19 to 21, 1938. On February 21 larvae were removed from log C1 and exposed to low temperatures, and log C2 was returned to normal winter temperatures at the log cache. On March 7 the log was returned to the laboratory and the cold-hardiness of the larvae determined.

Table 13 - Two day exposure to activity temperatures

Exposure	Log C1 Percent mortality	Log C2 Percent mortality
Normal larval resistance on Feb. 19, 1938		6% at -20°F
20°F	0	-
15°F	8	0
10°F	30	10
5°F	28	14
0°F	74	26
-5°F	82	46
-10°F	90	96
-15°F	98	94
-20°F	-	94
-25°F	-	98

These data show that a 2-day exposure to laboratory temperatures reduced larval resistance from a normal 6% mortality at -20°F to 98% at -15°F. It will be seen that the normal resistance on February 19 was somewhat lower than that which existed on February 7. The larvae from log C2 showed some response to their return to winter temperatures at the log cache, although on March 7 the cold-hardiness of the larvae was still below the normal of that date of 56% mortality at -25° F.

It is obvious that with limitations the length of exposure to activity temperatures determines the degree to which the cold-hardiness of mountain pine beetle larvae is reduced. This point is illustrated in the following summary tabulation:

Table 14

Exposure	Exposure to activity temperatures: 7 days	3 days	2 days
	Normal larval cold-hardiness: 0 mortality at -25°F	0 mortality at -25°F	6% mortality at -20°F
20°F	6% mortality	0% mortality	0% mortality
15°F	38% "	4% "	8% "
10°F	96% "	32% "	30% "
5°F	94% "	40% "	28% "
0	98% "	66% "	74% "
-5°F	100% "	96% "	82% "
-10°F		98% "	90% "
-15°F		100% "	98% "
-20 F			
-25°F			

These experiments failed to establish the length of exposure to activity temperatures as a factor in the ability of mountain pine beetle larvae to reestablish a broken cold-hardiness upon being returned to

normal winter temperatures. It is appreciated that subjecting active or non-resistant larvae to constant below-activity temperatures would not permit the development of much additional resistance, although it is believed that if such larvae were exposed to what might be normal hardening temperatures, such a relationship would exist. The lack of correlation between the larvae from the logs returned to the cache for the reestablishment of cold-hardiness no doubt resulted from the uneven air temperatures which existed in the field during that period.

As expected, these tests show that the cold-hardiness of mountain pine beetle larvae is not seasonal and can be broken at any time by exposing infested material to activity temperatures. The degree to which the resistance is reduced is governed by the duration and severity of such unseasonal activity temperatures. This indicates the possibility of unseasonal activity temperatures breaking the normal cold-hardiness of overwintering bark beetle larvae. It is believed that if such periods of warm weather were followed immediately by normal winter temperatures, severe brood mortalities would follow. Perhaps the rather common occurrence of brood mortality in the upper portion of infested trees may result from this combination of temperatures.

LABORATORY TECHNIQUES

Of the laboratory techniques necessary in conducting experiments with larvae in petri dishes to determine the cold-hardiness of mountain pine beetle larvae, the manner in which the larvae were handled before and after each experiment was perhaps of greatest importance. To preserve the existing status of cold-hardiness it was essential that the larvae be removed from infested logs as quickly as possible and under temperatures comparable to field conditions. Chilled petri dishes were used to prevent exposures to abnormal conditions.

As with some experiments it was necessary to hold petri dishes of larvae for periods of several days before using, a number of methods were tested. As moisture was considered as an important factor governing the successful storage of larvae at below-activity temperatures the following tests were conducted:

Test No. I: Eight petri dishes of 50 larvae each and an open vessel of water were placed in a covered container which was held at 35 to 40° F.

A. Four dishes were covered.

B. Four dishes left uncovered.

Test No. II: Eight covered petri dishes of 50 larvae each were placed on shelf in refrigerator and held at 35 to 40° F.

A. Four dishes had small (1/2 inch square) pieces of saturated blotting paper fastened to inside of lid.

B. Four dishes had no blotting paper.

Table 15 - Mortality of mountain pine beetle larvae stored in refrigerator 38°F - 40°F

Test	Dish	Percent mortality after 5 Days	10 Days	15 Days	30 Days	60 Days	109 Days	161 Days	Remarks
Covered petri dishes in covered container containing an open vessel of water									
									Larvae remained in fair
	1	0	0	0	0	0	6	20	condition. After 100
IA	2	0	0	0	0	0	0	4	days considerable mold
	3	0	0	0	Discontinued after				developed in these dishes.
	4	0	0	0	15 days, as petri				
					dishes were required				
					for other experi-				
					ments.				
Uncovered petri dishes in covered container containing an open vessel of water									
									Larvae remained in excel-
	1	0	0	0	0	0	0	18	lent condition. Mortality
IB	2	0	0	0	0	0	0	32	is believed to have resulted
	3	0	0	0	Discontinued after				from a condensation of
	4	0	0	0	15 days.				moisture which filled some
									of the larval cells.
Covered petri dishes on refrigerator shelf with small piece of saturated blotting paper in cover									
									Though there was little
	1	0	0	0	Discontinued				mortality in this test, the
IIA	2	0	0	0	0	0	0		larvae did not appear to be
	3	0	0	0	0	0	4		in as good condition as those
	4	0	0	0	Discontinued				in I. Larvae were decidedly
									drier and were quite yellow-
									ish in color.
Covered petri dishes on refrigerator shelf with no added moisture									
									Higher mortality and those
	1	0	0	0	Discontinued				surviving at 109 days were
IIB	2	0	0	0	6	14	70		quite dry and yellowish in
	3	0	0	0	4	26	78		color.
	4	0	0	0	Discontinued				

These tests indicate that moisture is an essential factor in the holding of mountain pine beetle larvae for extended periods, but for periods of 3 or 4 days any of the procedures would seem satisfactory. Furthermore, if proper moisture conditions are provided, it is possible that larvae could be stored for indefinite periods with no ill effects. Subsequent exposure tests showed that when resistant larvae were stored at below-activity temperatures there was no change in their original resistance or cold-hardiness.

When larvae are removed from an exposure to low temperatures many of them are frozen solid and turn white when exposed to the warmer air. As under field conditions such larvae would be subjected to a gradual change of temperatures, an attempt was made to duplicate this condition in the laboratory. Upon removal from the low-temperature cabinet the petri dishes of larvae were placed in petri dish holders and held in a large thermo jug of cracked ice. This procedure permitted a gradual warming of the larvae as the ice melted. The procedure proved quite successful and showed a higher survival than when check dishes of larvae were exposed to laboratory temperatures upon removal from the cabinet.

PREDICTION OF COLD-HARDINESS OF MOUNTAIN PINE BEETLE LARVAE

It is apparent that the cold-hardiness of mountain pine beetle larvae, which varies for different seasons, develops with the occurrence of below-activity temperatures in the fall of the year. In this development it is logical to assume that the degree of resistance is closely associated with the character of these so-called hardening temperatures. Although an unsuccessful attempt has been made to correlate established records of larval resistance with fall temperatures, it is possible that this failure rests in the character of temperature data which are available.

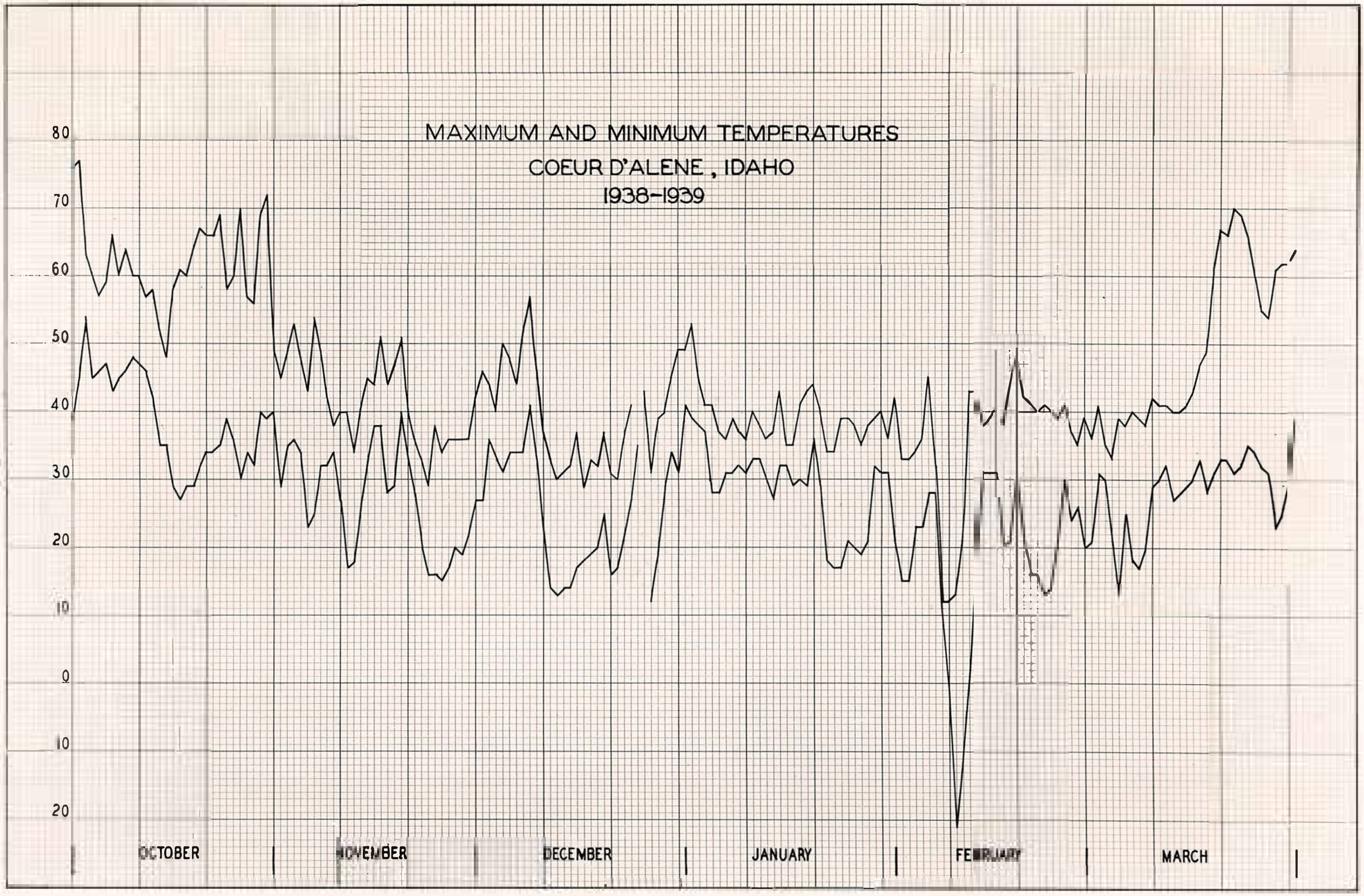
Maximum and minimum temperatures for the winters of 1938 to 1942 are shown in the following four charts. As these data depict weather conditions at Coeur d'Alene, Idaho, they only indicate a trend of temperatures existing at the log cache during these periods, which in most instances would be several degrees lower, especially in maximum reading.

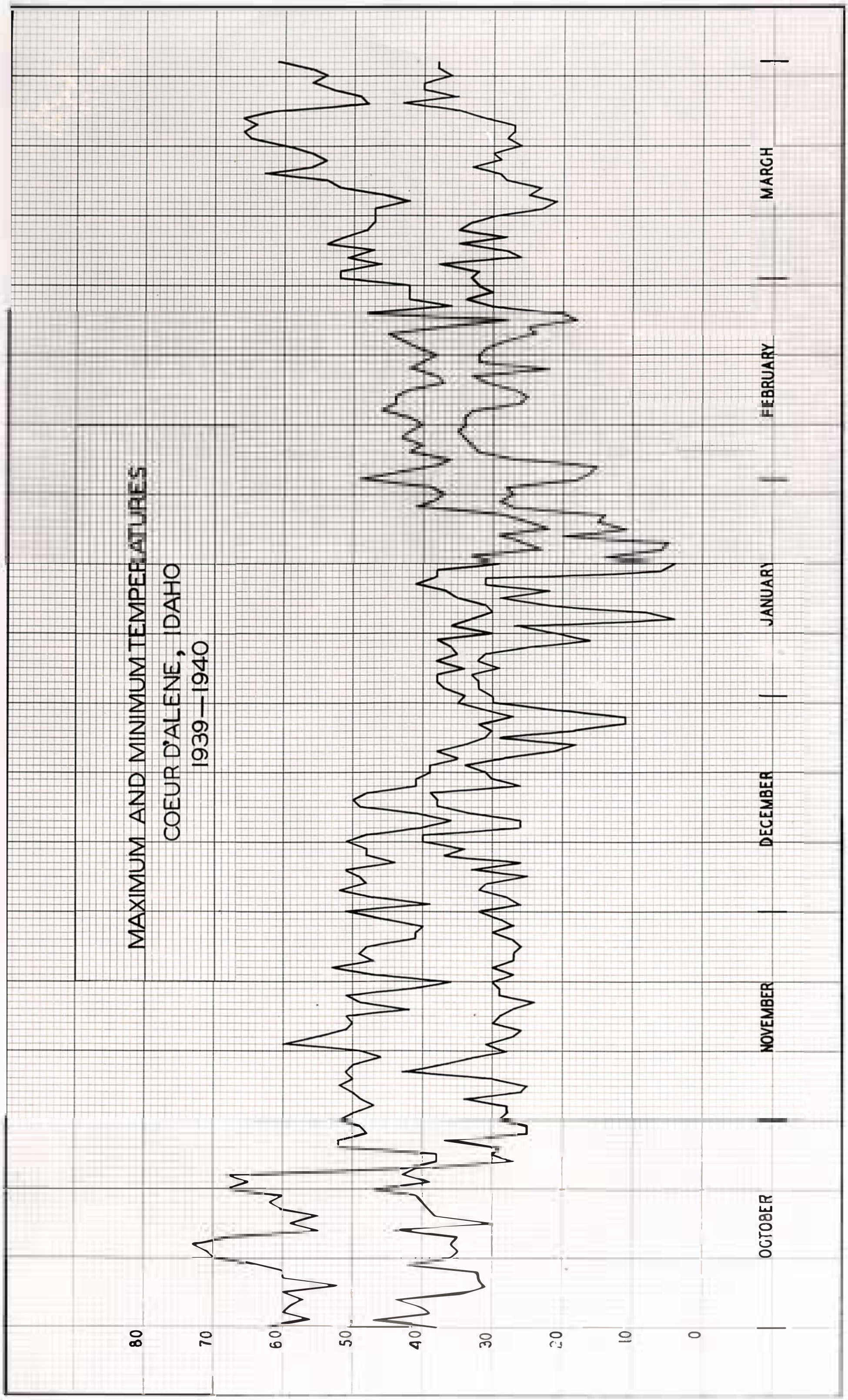
The variable cold-hardiness of larvae used during the different experiments is shown in the following tabulation:

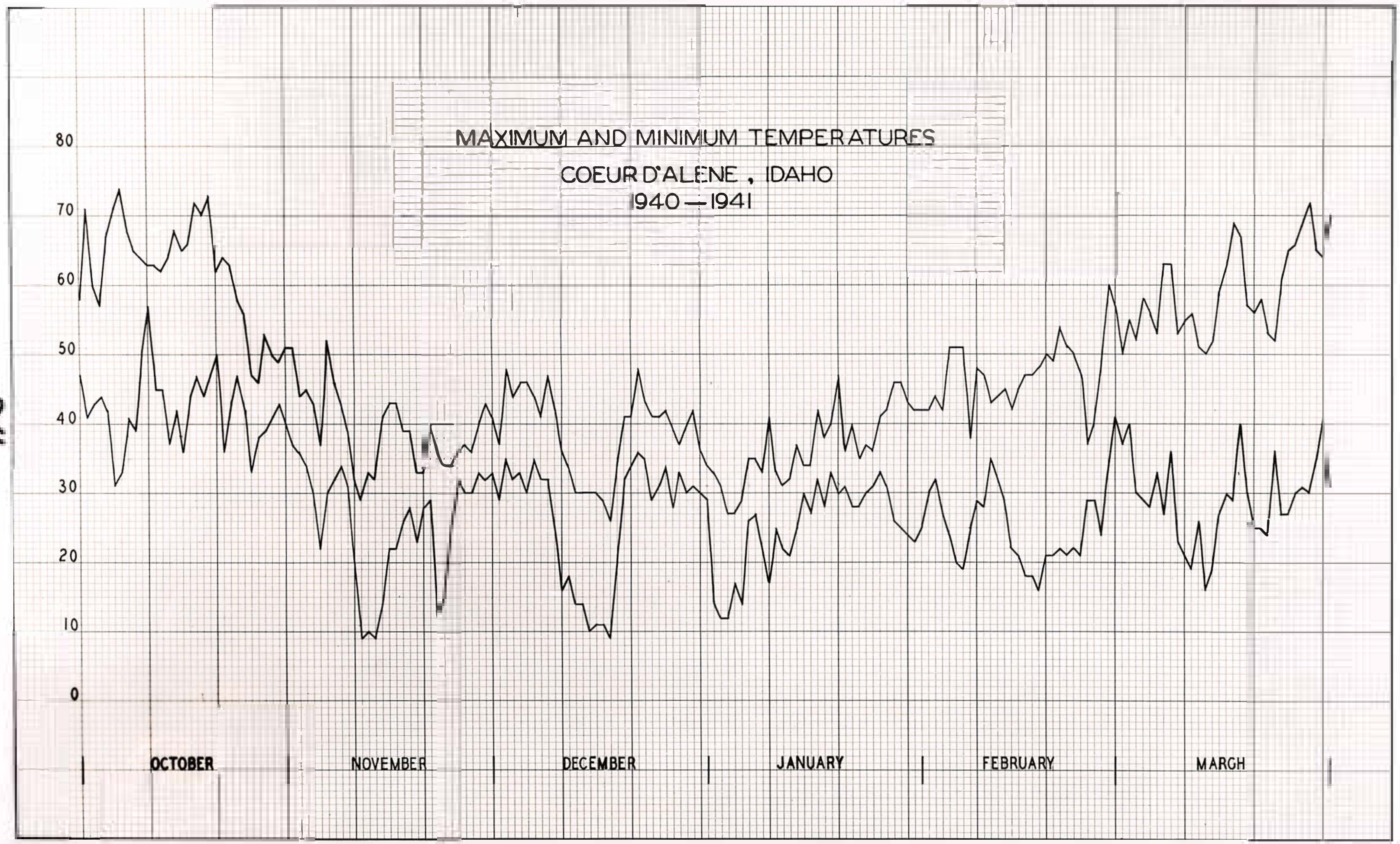
Table 16 - Yearly variations in the maximum cold-hardiness of mountain pine beetle larvae

Date	Brood mortality at $-30^{\circ}F$			
	Winter of 1938-39	Winter of 1939-40	Winter of 1940-41	Winter of 1941-42
December	15%	99%	100%	100%
January	9%	99%	31%	48%
February	0	97%	95%	94%
March	30%	100%	* 100%	100%

* A 100 percent mortality followed an exposure to $-15^{\circ}F$ in March 1941.

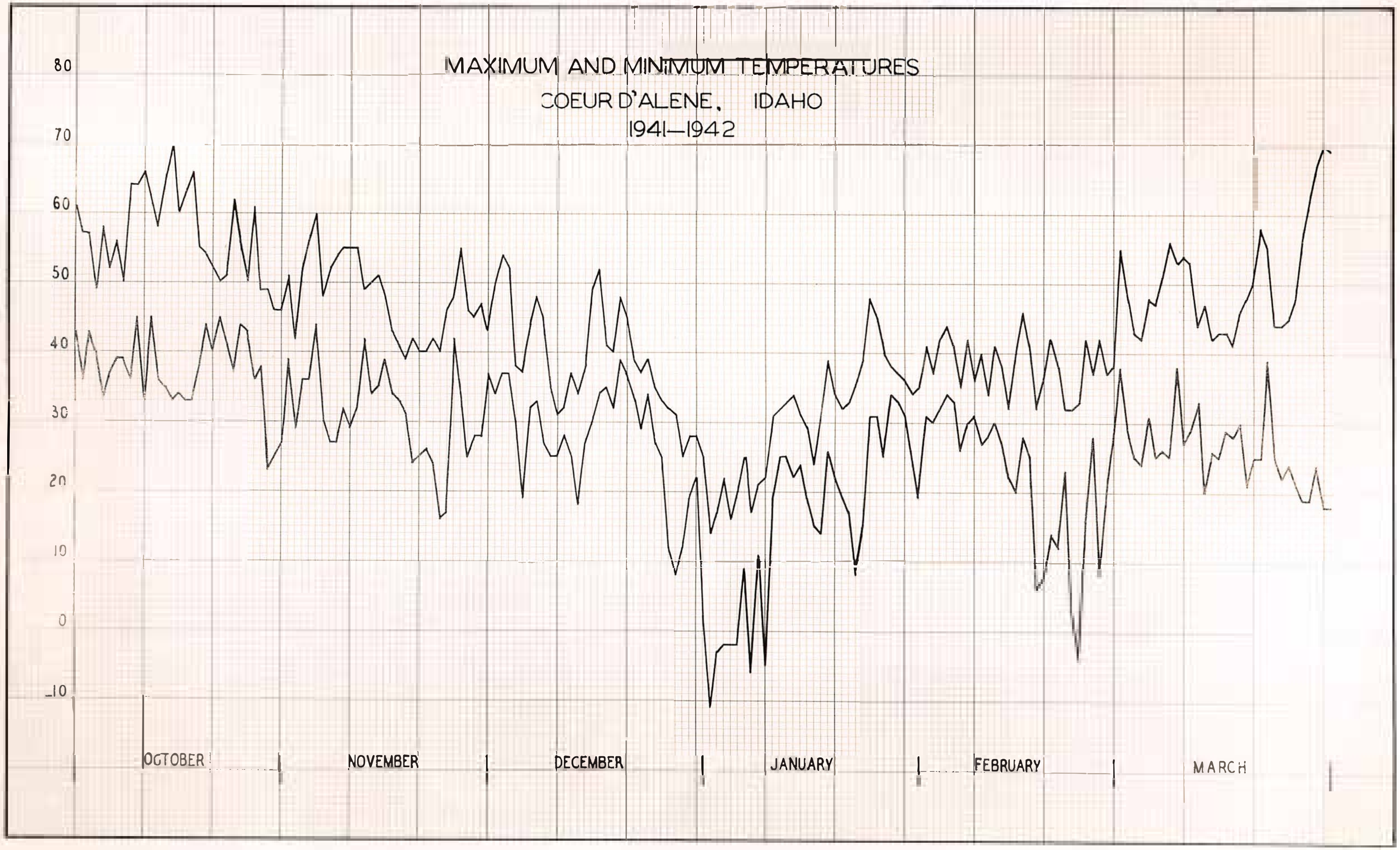

MAXIMUM AND MINIMUM TEMPERATURES
COEUR D'ALENE, IDAHO
1938-1939
80
70
60
50
40
30
20
10
0
10
20
OCTOBER
NOVEMBER
DECEMBER
JANUARY
FEBRUARY
MARCH


MAXIMUM AND MINIMUM TEMPERATURES
COEUR D'ALENE, IDAHO
1939—1940
80
70
60
50
40
30
20
10
0
OCTOBER
NOVEMBER
DECEMBER
JANUARY
FEBRUARY
MARCH



MAXIMUM AND MINIMUM TEMPERATURES
COEUR D'ALENE, IDAHO
1940—1941
80
70
60
50
40
30
20
10
0
OCTOBER
NOVEMBER
DECEMBER
JANUARY
FEBRUARY
MARCH

35


MAXIMUM AND MINIMUM TEMPERATURES
COEUR D'ALENE, IDAHO
1941—1942
80
70
60
50
40
30
20
10
0
-10
OCTOBER
NOVEMBER
DECEMBER
JANUARY
FEBRUARY
MARCH

This tabulation shows that a high degree (0 mortality at -30°F) of larval resistance existed during the winter of 1938-39. The following winter a much lower resistance (97% mortality at -30°F) occurred, and while the winters of 1940-41 and 1941-42 showed an increased resistance (31% and 48% respectively at -30°F), it was still much lower than that of the 1938-39 season.

In contemplating what might be considered as ideal temperatures for the development of cold-hardiness, it would seem that gradual increase in the severity of the weather would be most beneficial. Although the weather data fail to show such a correlation in the severity of the minimum temperatures and resistance, it would seem that maximum temperatures may be of greater importance. This belief is based upon the possibility that if below-activity minimums are followed by above-activity maximums, little if any larval cold-hardiness would be developed. On the other hand, if the larvae are subjected to below-activity temperatures (both minimum and maximum) for long periods, the severity of which is increased to that of severe winter conditions, it is believed that a high degree of cold-hardiness will follow. It will be seen that in 1938-39 the spread between maximum and minimum reading was much less than during the 1939-40 season. In November 1938 there was a total of 445 temperature degrees between the maximum and minimum reading, while in 1939 there were 653, or an increase of 47%. This fact would have little significance unless maximum temperatures were sufficiently low to arrest activity and to permit the processes of cold-hardening to continue. For example, in November 1938 the temperature rose above 50°F on only four occasions, with a total of 9 degrees. In 1939 there were 10 days of above 50°F temperature, with a total of 28 degrees. This analysis will not be carried further, for regardless of what correlation one might show, the data would have little significance, as the temperature records do not depict conditions within the area where cold-hardiness was developed. However, these data have been offered to illustrate the difficulty of predicting cold-hardiness, as well as the need for a more positive evaluation of the conditions which contribute to the development of cold-hardiness in larvae of the mountain pine beetle.

CONCLUSIONS

Conclusions drawn from this study do not appear to compliment the large volume of work conducted. Data obtained early in the course of this work depicted the same results as those finally drawn; however, repetitions of all tests under variable seasonal conditions were essential in establishing their significance. It was previously stated in this report that although there were still many points in connection with this study that had not been solved, it was believed that the practical demands had been satisfactorily answered. Perhaps the most important side phase of this study which has not been answered is the determination of the cold-hardiness of beneficial insects which tend to hold down the populations of the mountain pine beetle.

1. To meet the extreme temperature changes of a normal season mountain pine beetle larvae develop sufficient cold-hardiness to withstand the minimum low temperatures of the area. This development starts with the occurrence of cold weather in the fall of the year and increases in pace with the advent of winter conditions.

2. The occurrence of an abnormally low temperature at a time of year when a resistant cold-hardiness does not exist results in a mortality of mountain pine beetle larvae comparable to the severity of the temperature to which they have been exposed. Such unusual conditions can occur in the fall or early winter prior to the development of cold-hardiness or in the spring after larval resistance has been broken by activity temperatures.

3. Cold-hardiness can be broken by exposing the fully resistant larvae to activity temperatures. Obviously the length of this exposure will determine the extent to which the larval resistance to low temperatures is reduced. This fact opens the possibility that a period of abnormally warm weather could break the developed cold-hardiness of bark beetle larvae, which would render them susceptible to subsequent temperatures which under normal conditions would be non-lethal. The not uncommon mortality which occurs during the winter in the upper bole of infested trees is more likely caused by this combination of temperatures than by minimum low temperatures.

4. A relatively short exposure to a lethal temperature is as effective in producing mortality as a long one. Furthermore, a long exposure to a near lethal temperature is no more effective than a short one. Exposures of several days produced no higher larval mortality than one of two hours.

5. Mature larvae and pupae are more resistant to low temperatures than immature larvae.

6. At no time do all larvae in the same tree even under the same square foot of bark surface and in the same stage of development obtain the same degree of cold-hardiness.

7. Although larvae may be frozen solid, it is no assurance of mortality, as they often effect at least a temporary recovery.

8. Using infested logs in lieu of exposed larvae in petri dishes eliminates the factors of disturbance from normal winter hibernation and provides an accurate method of determining the mortality obtained from each experiment.

9. There is a distinct lag between subcortical and air temperatures. The thickness and character of the bark as well as the speed at which the air temperatures drop determine the extent of this lag.

10. The size of the log has no influence upon the lag between subcortical and air temperatures aside from the fact that thicker bark is expected on larger trees.